

OXIDASES IN HEALTHY AND IN CURLY-DWARF POTATOES

By H. H. BUNZEL,

*Chemical Biologist, Plant Physiological and Fermentation Investigations,
Bureau of Plant Industry*

INTRODUCTION

The curly-dwarf, among other related potato diseases, has been described very fully in one of the recent publications of this Department (Orton, 1914),¹ in which the very confusing literature on the subject of potato maladies referred to vaguely in the past as leaf-roll, curly-top, blight, Kräuselkrankheit, Blattrollkrankheit, etc., is critically reviewed. On the basis of this review and of the work done by the Office of Cotton and Truck Diseases and Sugar-Beet Investigations, which has made a thorough survey of the principal potato districts on this continent, as well as abroad, a number of distinct diseases are recognized, each with its characteristic symptoms and probable cause.

Some of these diseases, particularly the leaf-roll and the curly-dwarf, can not be traced to organisms of any sort for their origin and are supposedly disturbances of a purely physiological nature. To throw light on this matter, Mr. W. A. Orton, of the Bureau of Plant Industry, requested the writer to make a quantitative study of the oxidizing enzymes of potatoes at Houlton, Me., and immediate vicinity. Oxidase determinations were there carried out with healthy material, as well as with plants having the curly-dwarf disease. In this paper only such plants were considered to have curly-dwarf as showed the characteristic symptoms described by Orton (1914).

This is not the first attempt to correlate enzymatic disturbances with plant diseases. Sorauer (1908) was the first one to attribute the leaf-roll of potatoes to disturbances in the oxidase mechanism of the tubers. According to this author, the dark patches observable on the cut surfaces of such tubers are due to a greater oxidase content than is found in normal tubers; the abnormalities of the foliage are due to malnutrition through the tubers. His conclusions are based on chemical experiments of Grüss (1907). The most important and complete investigation of the subject was made by Doby (1911-12), who reached the very important conclusion that the oxidase content of the diseased tubers is greater than that of the normal ones. He also found a higher ash content and lower percentage of starch and insoluble protein in the diseased tubers, stating

¹ Bibliographic citations in parentheses refer to "Literature cited," p. 403-404.

that the increased ash is probably responsible for the increased oxidase activity, judging from the work of Bertrand (1897) and others (Dony-Hénault, 1908; Dony-Hénault and Van Duuren, 1907; Trillat, 1903 and 1904). The diminution of the starch and protein in the diseased tubers is the net result of the increased combustion of their cleavage products. Doby (1911-12) measured the oxidation of pyrogallol according to the method of Bach and Chodat (1904), with and without the addition of hydrogen peroxid, and also measured the oxidation of tyrosin according to an optical method (König and Krüss, 1904). In general, the peroxidase, oxygenase, and tyrosinase were present in larger quantities in the curly-dwarf tubers than in the normal ones.

With the manometric method devised by the writer, it was found that in sugar beets affected with curly-top the oxidase content of the foliage is two to three times as great as that of normal beet leaves. This same difference was found to exist when plants were studied whose growth had been retarded by causes other than the curly-top (1912).

DESCRIPTION OF EXPERIMENTAL METHODS

For all of the experiments with leaves, fresh material was used—that is, plants collected on the day of experimentation. In the case of tubers it was sometimes necessary to allow them to lie for a few days until enough material for a complete series of experiments had accumulated. During the whole period of 10 weeks the plants were collected in the field at 7.15 a. m. and taken to the laboratory at once. The weight of the foliage was there determined and the material then ground up in a meat chopper. The juice was pressed out of the pulp by hand through a silk cloth.

Nearly all of the experiments with normal material were made on plants of one variety, the Green Mountain. With one or two intentional exceptions the samples of normal plants were taken from the same field. They were grown under fairly uniform conditions of environment, and the soil was fertilized uniformly with the same fertilizer. All of the pathological material was collected on a field several miles away, necessarily from plants of different varieties, but all grown on the same type of soil and with the same kind of fertilizer as that used in the field on which the normal plants were collected. In most cases 25-gram samples of the juice were preserved with about 100 c. c. of 25 per cent alcohol, in order that the solid contents of the juices examined might be determined, in case it seemed necessary.

The experiments were carried out in the same manner as that described in a former publication (Bunzel, 1912). The following 18 ring compounds were used as reagents to determine the oxidase activity of the juices: Benzidin, pyrogallol, alphanaphthol, leuco base of malachite green, phloroglucin, aloin, pyrocatechol, tyrosin, hydrochinone, phloridzin,

resorcin, guaiacol, orthocresol, metacresol, paracresol, orthotoluidin, metatoluidin, and paratoluidin.

In the case of each of the solid substances 0.05 gram was weighed out for each determination. In the experiments with guaiacol 4 drops (0.15 gram) were used; by separate experiments it was shown that this quantity gave the highest result under the conditions of the experiments. The cresols and toluidins were found to be very poisonous, inhibiting the action of the potato oxidases when used in too great quantities. By a series of experiments it was found that 2 drops of each gave the optimum result. As in previous experiments, 1 c. c. of normal sodium hydrate was used in the glass basket in all experiments with pyrogallol, to absorb the carbon dioxide produced during the oxidation.

All of the experiments described herein were carried out at 41° C. The apparatus used were all of the small type, in which a change in pressure of 1 cm. of mercury corresponds to the absorption of 1 c. c. of oxygen. The rate of shaking was 5 complete excursions in 3 seconds. All of the results were expressed in terms of the oxidase unit previously used by the writer. The unit is an oxidase solution of such strength that 1 liter of it can bring about the oxidation of the equivalent of 1 gram of hydrogen (1912, p. 40). Blank determinations with the reagents here used showed that no measurable oxidase absorption took place under the conditions of the experiments in the absence of plant juice.

The thermostat box was provided with a false bottom about 6 inches above the floor of the box and a free space of 4 inches at each end for the sake of free circulation. The heating lamps were all arranged below this false bottom. In this way very uniform heating throughout was attained. The stopcocks were closed through an opening just large enough to admit the arm, instead of opening a window, as was done formerly. To reduce still more the disturbances of temperature within the box, the opening for the arm was protected by means of a sleeve into which the arm was slipped.

Although the results obtained with the method here used are more accurate and reliable than those obtained with any other existing method, yet this method is not entirely free from sources of error. It is probable that a part of the oxidases are destroyed by the shaking at the comparatively high temperature (41° C.). It is also probable that the reagents used for the oxidation act as poisons even in the small concentrations in which they are present. Probably, however, it will be only a matter of time before these possible sources of errors will be eliminated. For the present it may be said that the results were obtained in experiments carried out under identical conditions and are therefore comparable. In all the experiments the juice was pressed out of the ground pulp by hand and by the same operator. While it may seem that juices of more uniform composition might be obtained by pressing them out with a

machine, separate experiments show that a portion of the activity is lost thereby. The fresh hand-pressed potato juice had an activity of 0.287 units (pyrogallol), while the juice pressed out of the remaining pulp by means of a hydraulic press at a pressure up to 15 tons on a 6-inch circular ram was 0.170 units (pyrogallol) and the juice pressed out at a still higher pressure had an activity of only 0.107 units (pyrogallol). Inasmuch as the juice obtained by means of the hydraulic press had to pass through an appreciable amount of compressed pulp, it is probable that the diminished activity of the machine-pressed juice was due to the loss of a part of the oxidases by adsorption.

In this connection the results of Dixon and Atkins (1913) are very interesting. They found that in successive pressings of leaves (*Hedera helix*) in a vise, juices with increasing concentration of electrolytes were obtained. They experimented also with leaves treated with liquid air and concluded that the only way to obtain juices corresponding to the concentration of the sap in the vacuoles of the uninjured tissues is to press them out after exposure to liquid air. Unfortunately, such procedure was impossible during this work, which had to be carried out in the field.

RATE OF GROWTH OF THE POTATO PLANT

In former publications it has been shown that the oxidase content of juices bears a very definite relation to the rate of development of the particular plant specimens from which they are derived. In the sugar beet, which the writer studied in this respect, the oxidase content of the foliage runs up appreciably when the normal growth of the plants is interfered with by drought, excessive watering, diseases, etc. In the foliage of normally developing sugar-beet plants the oxidase content of the juice is only about one-half that of stunted plants. On the basis of the results obtained with sugar beets the following generalizations can be made:

- Normal growth.....Normal (low) oxidase content.
- Retarded growth.....Abnormal (high) oxidase content.

The recognition of this fact led to an examination of the rate of growth of the potato plants which were used in this research. Table I shows the relation which the size of the shoots and the foliage of all the plants in a hill, as well as of the single shoots, bears to the age of the plants.

TABLE I.—Relation of the total weight of the shoots of the whole hills, as well as of the single shoots, to the age of the potato plants

Series No.	Date of collection.	Age.	Total weight of shoots.	Number of hills.	Mean total weight of shoots per hill.	Number of shoots.	Mean weight of shoots.
		Days.	Grams.		Grams.		Grams.
1.	July 9	29	84	5	17
2.	July 10	30	102	7	15	17	6
4.	July 11	31	150	8	19	22	6.8
6.	July 12	32	108	8	13.5	19	5.7
9.	July 14	34	222	15	15	32	6.9
10.	July 15	35	297	9	33	23	12.9
12.	July 31	19	75	7	11	40	1.9
14.	July 31	19	100	6	17	41	2.4
15.	Aug. 1	20	170	6	28	64	2.7
18.	Aug. 2	60	360	1	360	3	120
21.	Aug. 4	62	750	2	375	4	188
24.	Aug. 8	66	680	1	680	4	170
26.	Aug. 9	67	368	1	368	1	368
29.	Aug. 11	30	365	5	73	44	8.3
32.	Aug. 21	40	500	1	500	7	71.4
35.	Aug. 29	88	350	1	350	1	350
38.	Sept. 8	98	375	1	375	1	375

In order to present these data more clearly, they were plotted as shown in figure 1. The ages of the plants are measured off on the abscissæ and the weight of the shoots on the ordinates. The continuous line corresponds to the development of the plants (the total weight of the shoots of one hill), and the broken line corresponds to the mean rate of development of all of the single shoots of one hill.

The irregularities of the curve representing the growth of the shoots of a whole hill are apparently due to variations in the number of stalks contained therein. This becomes strikingly apparent from the smoothness of the curve representing the growth of single stalks. With practically no interruption this curve shows a gradual increase in size until the sixty-seventh day is reached, growth of the stalks apparently stopping at that point. The curve from this point on is practically a straight line.

OXIDASES OF HEALTHY POTATO PLANTS

In order to be able to compare the oxidase activities of diseased potato plants with healthy ones at the same stage of development, it was essential to establish the oxidase content of healthy material at all stages of development. Such a study on normal plants is also of general physiological interest. While the excellent work of Palladin (1906) and his school has shown that the respiration of plants takes place in stages corresponding to several distinct respiratory enzymes, they have made no measurements of the oxidizing power of these respiratory enzymes. Moreover, working with frozen wheat seedlings and those not frozen and with etiolated leaves of *Vicia faba* and leaves of *Plectogyne japonica*, they con-

clude that "oxidase" and "oxygenase" are practically lacking in embryonic organs and that their concentration in these plants rises during growth and diminishes again when growth has stopped. They draw their conclusions entirely from the quantity of CO_2 liberated by the

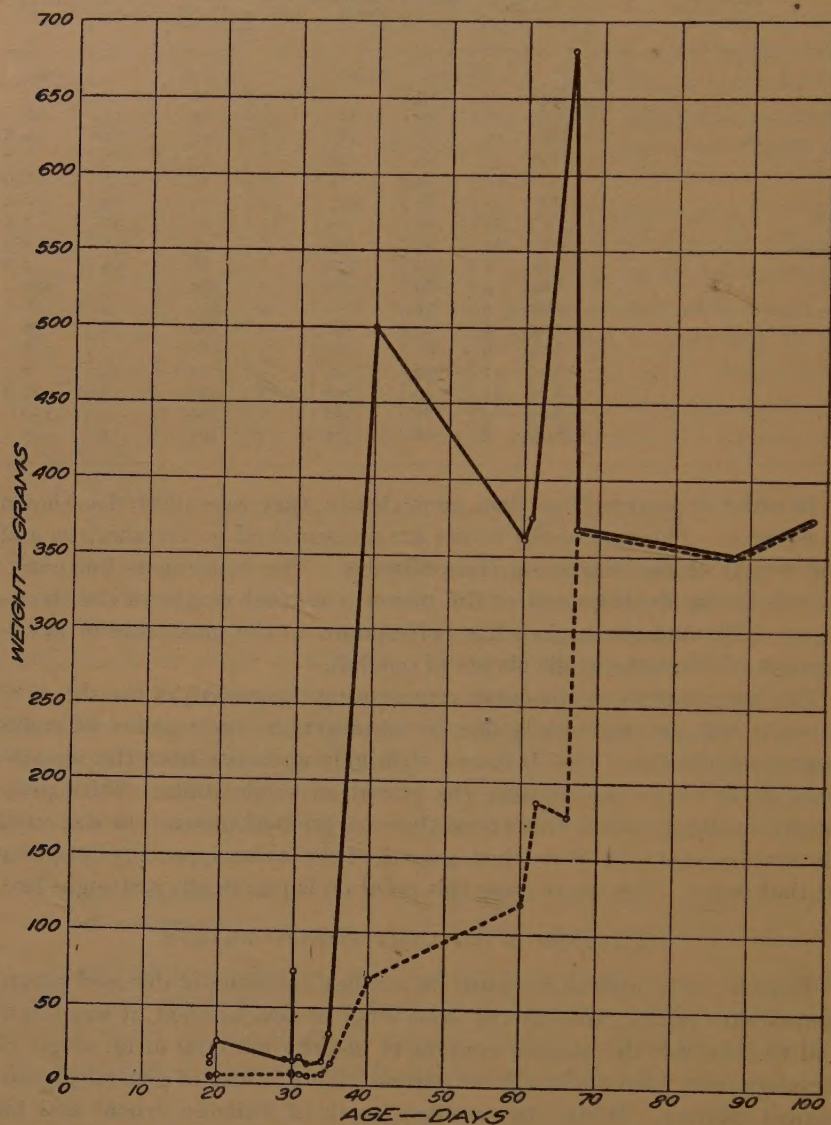


FIG. 1.—Rate of development of the aerial portion of potato plants and mean rate of development of all the single shoots in one hill.

plant organs under different conditions. It seemed of great interest, therefore, to find out what relation the concentrations of the oxidases present in the pressed-out sap of a plant bear to the state of development of the same plant.

OXIDASE ACTIVITY OF THE JUICE OF THE SHOOTS

The results obtained in the measurement of the oxidases in the juice of healthy potato plants of the same variety at various ages, grown under normal and as nearly identical conditions as possible, are given in Tables II to VIII, and some of the results are also shown graphically in figures 2 to 20.

The shoots were taken from the plants

immediately above the point where they emerged from the soil. In figures 2 to 5¹ the abscissæ represent the age of the plants as measured from the time of planting, and the ordinates the activities of the juices as measured in the oxidation of the various aromatic compounds used. These data show a distinct downward tendency; there is apparently a marked diminution in the oxidase activities of the pressed-out juice of the shoots during the beginning of their growth.

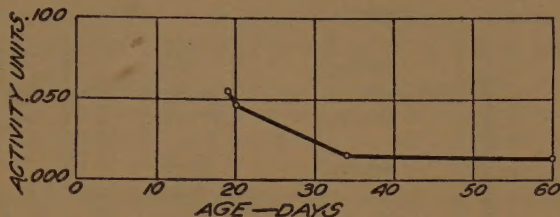


FIG. 2.—Curve showing oxidation of leuco base of malachite green in the presence of the juice of green potato shoots.

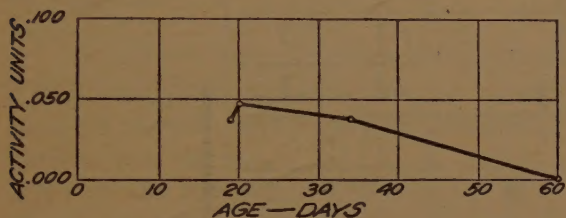


FIG. 3.—Curve showing oxidation of aloin in the presence of the juice of green potato shoots.

OXIDASE ACTIVITY OF THE JUICE OF THE STEMS

Experiments on sugar beets showed that the juice obtained from the stems of the plants exhibited very much less oxidase activity than that of the leaves (Bunzel, 1913a; 1913b). It seemed probable, therefore, that the juice of the stems of the potato plants examined would also be less active than the juice of the leaves. A comparative increase

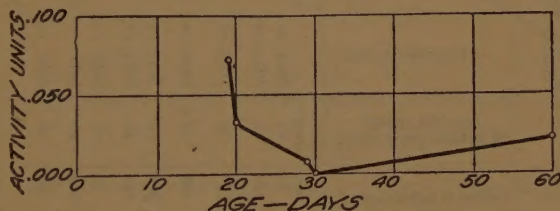


FIG. 4.—Curve showing oxidation of tyrosin in the presence of the juice of green potato shoots.

¹ Inasmuch as nearly all of the curves obtained in working with the 18 reagents show the same general relationships, for the sake of a briefer and, therefore, more comprehensive presentation of the facts, only 4 to 8 curves are presented in the case of the shoots, foliage, and tubers, respectively. While these curves were not picked at random, they are typical of the situation in each case. The writer felt justified in doing this, inasmuch as all of the curves can be constructed from the tables in the text, and since the results are numerically compared in Table XII.

TABLE II.—*Oxidase activities of the juice of shoots of healthy potato plants*

Series No.	Date.	Number of hills used.	Total weight of shoots.		Mean total weight of shoots per hill.	Date of planting.	Age of plant.	Oxidase activity expressed in units as measured in the oxidation of the reagents.																																				
			Grams.	Grams.				Benzidin.	Pyrogallol.	α -naphthol.	L. b. of m. g.	Phloroglucin.	Alcin.	Pyrocatechol.	Tyrosin.	Hydrochinone.	Phloridzin.	Resorcin.	Guaiacol.	O-cresol.	M-cresol.	P-cresol.	O-toluidin.	M-toluidin.	P-toluidin.																			
1.....	July 9	5	84	17	June 10	29	Days.																				
2.....	July 10	7	102	15		30																					
3.....	July 11	8	150	19		31																				
4.....	July 12	8	108	13.5	June 10	32																				
5.....	July 14	15	222	15		34																					
6.....	July 15	9	297	33		35																				
7.....	July 31	7	75	11	July 12	19																				
8.....	July 31	6	100	17		19																					
9.....	Aug. 1	6	170	28		20																				
10.....	Aug. 2	1	360	360	June 3	60																				
11.....					
12.....					
13.....																				
14.....																				
15.....																				
16.....																				
17.....																				
18.....																				
19.....																				
20.....																				

^a Leuco base of malachite green.

in size of the stem of a growing plant as compared with the foliage or a diminution with age of the activity of the stem juice as compared with the activity of the foliage juice would therefore result in a diminution with age of the activity of the juice of the aboveground portion of the plant. Consequently oxidase determinations were made on the juice obtained from stems of plants 69 days old. The results are given in Table III.

TABLE III.—Oxidase activities of the juice of stems of plants 69 days old

Reagent.	Activity. ^a	Reagent.	Activity. ^a
Benzidin.....	0.035	Phloridzin.....	0.078
Pyrogallol.....	0	Resorcin.....	0
α -naphthol.....	0	Guaiacol.....	.023
Leuco base of malachite green.....	0	O-cresol.....	0
Aloin.....	.031	M-cresol.....	.101
Phloroglucin.....	0	P-cresol.....	.168
Pyrocatechol.....	.023	O-toluidin.....	0
Tyrosin.....	0	M-toluidin.....	0
Hydrochinone.....	0	P-toluidin.....	0

^a Activity expressed in units as measured in the oxidation of the reagents.

The stem juice proved to have no activity whatever toward 11 of these 18 reagents, and toward the remaining 7 it was slight in comparison with the activity of the foliage juice, as will be shown later. That the stem during growth increased in weight more rapidly than the remainder of the shoot is shown in Table IV (column 9). It is probable, therefore, that the diminishing activity of the juice of the shoots of the potato plant was due to increasing dilution with age of the very active leaf juice with the relatively inactive stem juice.

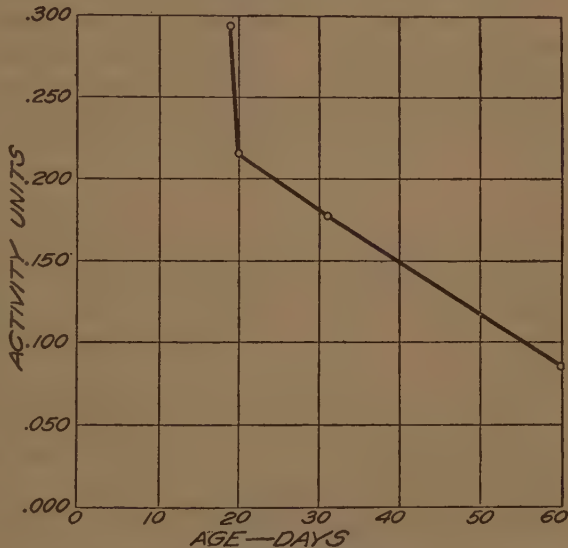


FIG. 5.—Curve showing oxidation of phloridzin in the presence of the juice of green potato shoots.

OXIDASE ACTIVITY OF THE JUICE OF THE LEAVES

The leaves proper are the seat of the greatest physiological activity in plants. The food of the plant is largely synthesized in the leaves and also in part broken down in them, according to the needs of the plant.

In physiological disturbances, such as the curly-dwarf disease of potatoes and the curly-top of sugar beets appear to be, the leaves are the parts primarily affected. It is therefore to be expected that any chemical differences existing between healthy plants and plants affected with the curly-dwarf disease will be most pronounced in the leaves proper. In order to be certain of results which represent the activity of the juice of

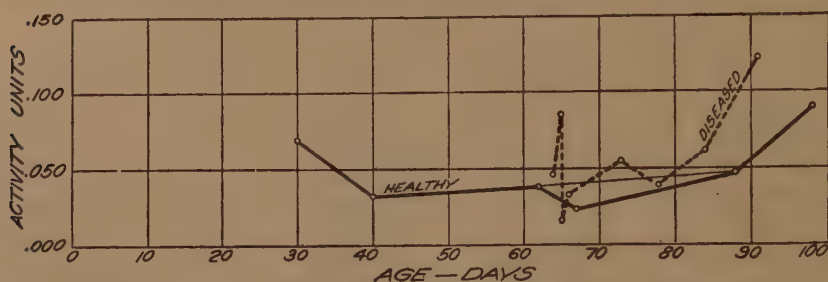


FIG. 6.—Curve showing oxidation of leuco base of malachite green in the presence of the juice of potato foliage.

the foliage proper, all experiments with the green parts of the potato plant were from this point on carried out on leaves alone. Table IV gives the data on the activity of the leaf juice alone.

Some of these results are also graphically presented in figures 6 to 12. (See footnote, p. 379.) For easy comparison the figures obtained with healthy leaves, as well as those obtained later with curly-dwarf leaves, were plotted on the same systems of coordinates. The continuous lines

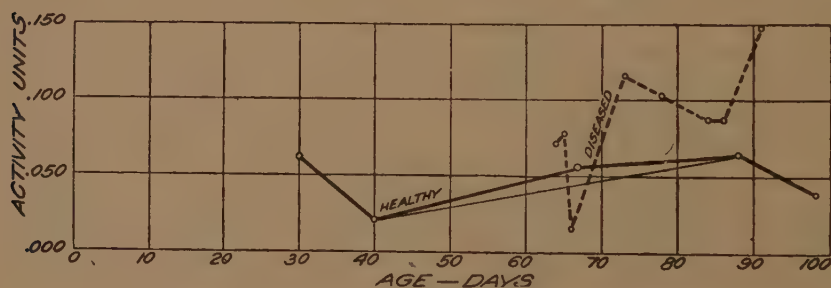


FIG. 7.—Curve showing oxidation of pyrocatechol in the presence of the juice of potato foliage.

represent the results obtained with healthy plants, the dotted lines those with the diseased ones.

These curves show great fluctuations of oxidase content. Barring some of the irregularities, probably due to individual peculiarities of the samples examined, the curves take a downward direction at first, remain at a low level for a prolonged period, and take an upward movement again towards the end. The lowest point in the curve is reached generally on the fortieth day; the period of low oxidase content extends to a point of time between the sixtieth and eightieth day, when the upward movement of

TABLE IV.—*Oxidase activities of the juice of the leaves of healthy potato plants*

Series No.	Date.	Number of hills used.	Total weight of shoots.	Mean total weight of shoots per hill.	Date of planting.	Age of plant.	Total weight of stems.	Stems × 100.	Oxidase activity expressed in units as measured in the oxidation of the reagents.																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																												
									Benzidin.	Pyrogallol.	α -naphthol.	I. b. of m. & a.	Phloroglucin.	Alcin.	Pyrocatechol.	Tyrosin.	Hydrochinone.	Phloridzin.	Resorcin.	Guaiacol.	O-cresol.	M-cresol.	P-cresol.	O-toluidin.	M-toluidin.	P-toluidin.																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																																											
21.....	Aug. 4	2	750	375	June 3	62	402	54	.055	.016	.035	.039	.035	.047043	.0456	.0109	.059	.059</

^a Leuco base of malachite green.

the curve generally begins. The juice from the plant collected on the sixty-seventh day seems unusually rich in oxidases. If the points obtained from the data on this plant were discarded, the curves would be all quite regular, with the exception of those corresponding to the oxidation of hydrochinone and of some of the cresols. To show what types of

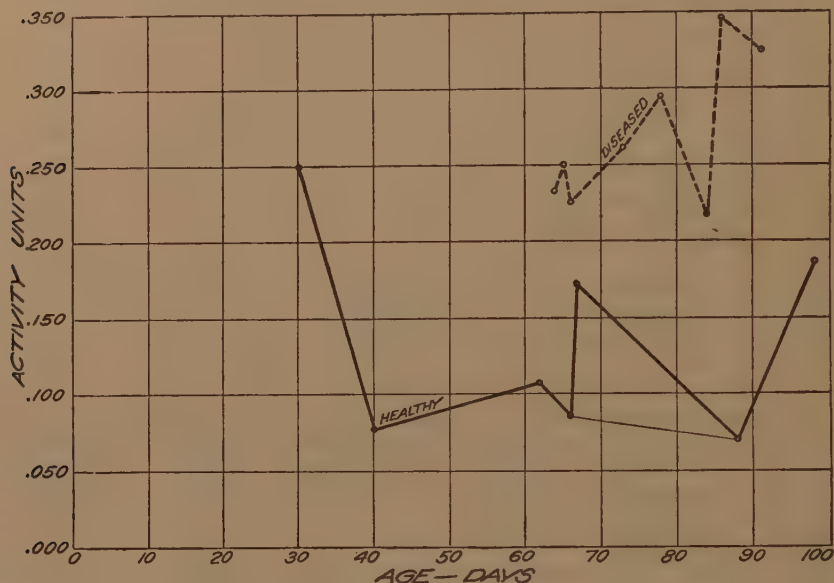


FIG. 8.—Curve showing oxidation of phloridzin in the presence of the juice of potato foliage.

curves would be obtained by elimination of the points obtained for the sixty-seventh day of growth, which point seems irregular, the adjacent points on both sides of the "sixty-seventh-day point" are connected with relatively thin lines to complete the curves; the initial fall and the final rise thus become very apparent and clear-cut.

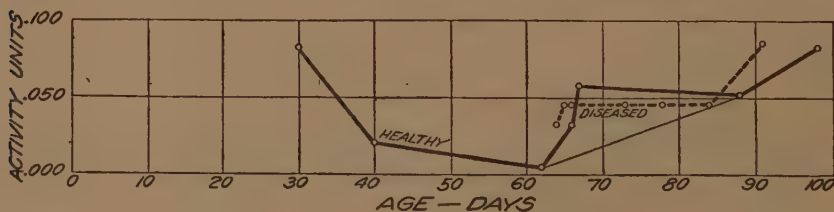


FIG. 9.—Curve showing oxidation of o-cresol in the presence of the juice of potato foliage.

The curves, of course, are not smooth. This is to be expected when it is considered that there are numerous factors influencing the physiological condition of the plants. Differences in the nature of the seed, of the soil, and many other factors probably influence the development of the plant qualitatively as well as quantitatively.

With all of the reagents except guaiacol and metacresol, the rise in the oxidase content observed during the second half of the period of examination coincides approximately with the stoppage of growth, which point is shown in figure 1 to be about the sixty-seventh day. In this respect, therefore, these results are in striking harmony with those obtained while working on diseased sugar beets. In the case of sugar beets the writer has shown that the factors which had a retarding influence on the growth of the plants also caused the oxidase content of the juice of their foliage

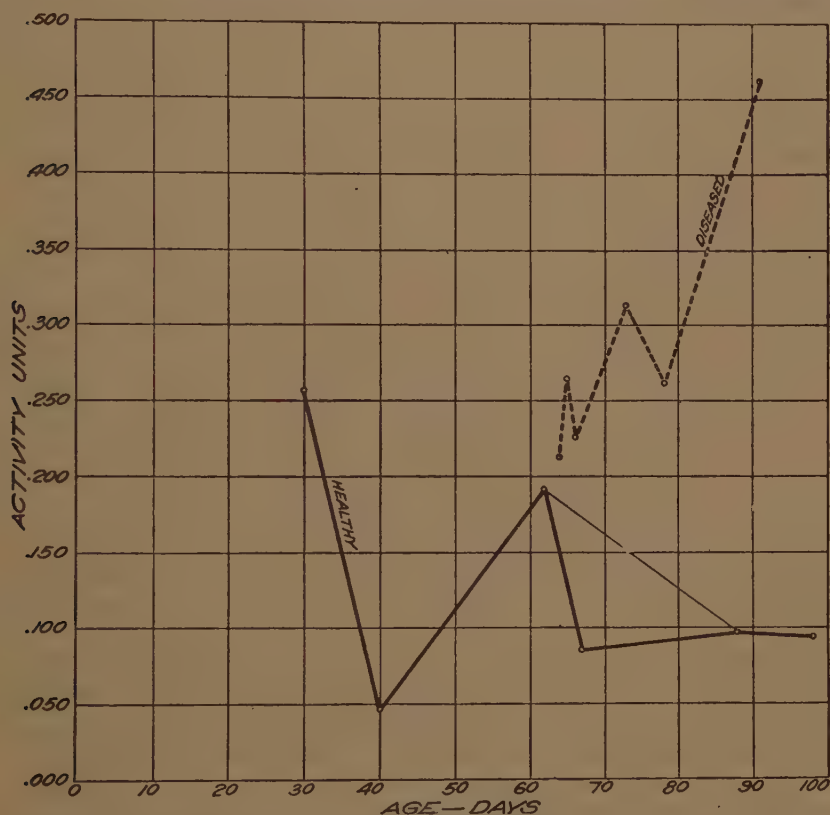


FIG. 10.—Curve showing oxidation of m-cresol in the presence of the juice of potato foliage.

to increase. It is possible that the same factors which led to an increased oxidase content during the retardation of growth of sugar beets will lead to a similar rise during normal cessation of growth in potato plants.

OXIDASE ACTIVITY OF THE JUICE OF THE SPROUTS AND OF THE TUBERS FROM WHICH THE SPROUTS HAD BEEN REMOVED

The relatively high oxidase content of very young potato plants suggested an examination of the sprouts from the tubers. Seed tubers of the Green Mountain variety of the same stock as was used for all of the

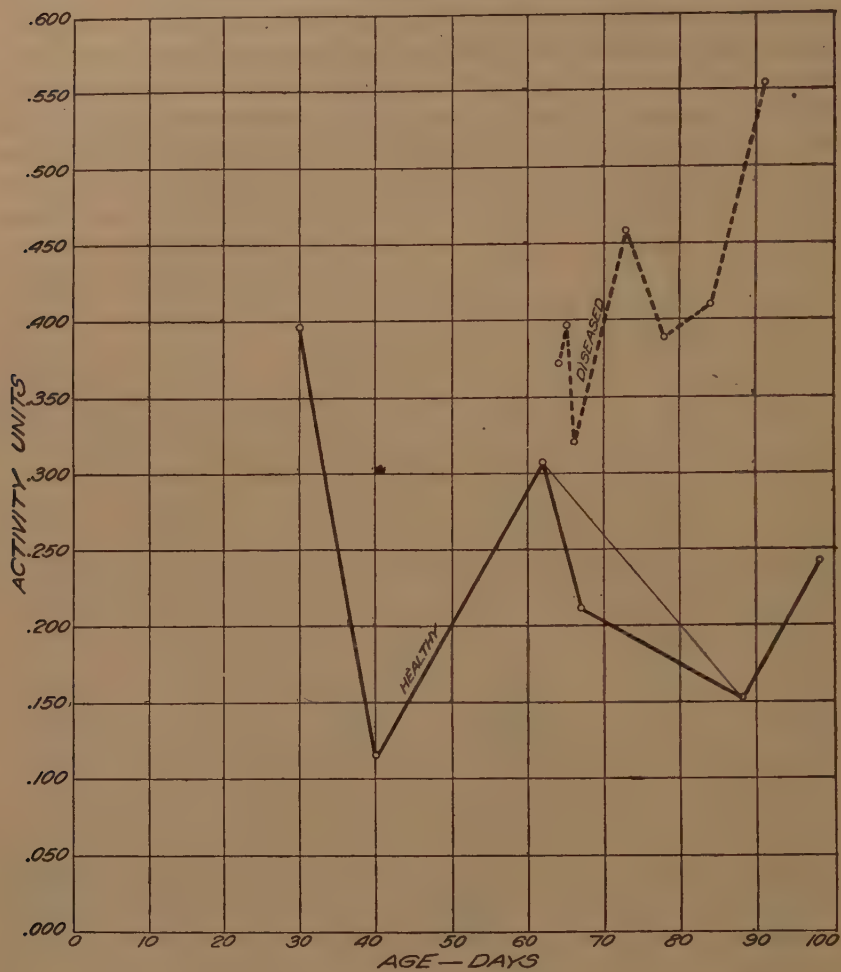


FIG. 11.—Curve showing oxidation of p-cresol in the presence of the juice of potato foliage.

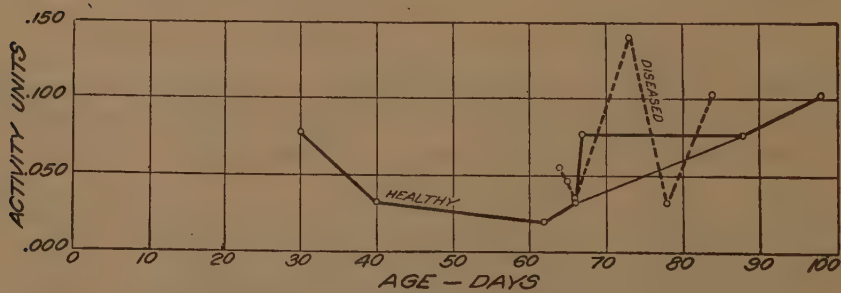


FIG. 12.—Curve showing oxidation of m-toluidin in the presence of the juice of potato foliage.

experiments already described in this paper were stored at room temperature from July 12 to September 3, 1913. During that time the tubers were lying on a table exposed to the light, and the temperature during the period fluctuated between 15° and 25° C. There were 36 tubers, soft but sound, yielding 170 grams of sprouts; the 36 tubers from which the sprouts had been removed weighed 2,670 grams. Only 22 c. c. of juice were obtained from the sprouts in the usual way. This juice turned immediately to a chocolate-brown color. The oxidase activity of the juice of the sprouts toward seven of the reagents is given in Table V. The oxidase activity of the juice of the tubers toward all of the reagents is given in Table VI.

TABLE V.—*Oxidase activities of the juice of the sprouts from Green Mountain seed-potato tubers*

Series No.	Oxidase activity expressed in units as measured in the oxidation of the reagents.					
	Pyrocatechol.	Tyrosin.	Phloridzin.	Resorcin.	Hydrochinone.	Paracresol.
41.....	0.562	0.296	0.359	0.055
42.....	0.807	0.488
						0.495

¹ Leuco base of malachite green.

OXIDASE ACTIVITY OF THE JUICE OF THE TUBERS

The method of procedure in the case of the potato tubers was the same as that used with the foliage. The tubers were freed from adhering soil by means of cold, running water and were wiped dry with a clean towel and ground up whole. It has been known for some time that the juice obtained from the layers of the potato tuber near the surface is more active than that from the inner portions. Notwithstanding this variation of oxidase activity in different parts of the tuber, the whole tubers, including the peel, were used for these experiments. This was done to avoid the introduction of new factors. The results are presented in Table VII.

To see whether the oxidase content of the juice from the tubers bears any relation to either the age of the plant from which the tubers are derived or their own weight, the ratio of oxidase content to age and weight was represented graphically. In the figures 13 to 20 (see footnote, p. 379) two sets of curves are given. In both sets the oxidase activities of the juices are shown on the ordinates, while the ages and weights, respectively, are laid off on the abscissæ. The curves formed by the continuous lines show the relation of age to oxidase content and the curves formed by the thin broken lines show the relation of the size of the tubers to their oxidase content.

From these curves no definite relationship is apparent between the oxidase content of the tubers on the one hand and their age or size on

TABLE VI.—Oxidase activities of the juice of healthy potato tubers which sprouted in the laboratory and from which the sprouts had been removed

Series No.	Oxidase activity expressed in units as measured in the oxidation of the reagents.										
	Benzidin.	Pyrogallol.	α -naphthol.	L. b. of m. g. glucin.	Phloroglucin.	Alolin.	Pyrocatechol.	Tyrosin.	Hydrochinone.	Phloridzin.	Resorcin.
43	0.332	0.203	0.140	0.012	0.047	0.113	0.413	0.250	0.710	0.273	0.023
44
45
										0	0.257
											0.952
											0
											0.012
											0.156

^a Leuco base of malachite green.

TABLE VII.—Oxidase activities of the juice of healthy potato tubers

Oxidase activity expressed in units as measured in the oxidation of the reagents.																														
Series No.	Series No. of leaves of same plant.	Date of collection.	Date of experiment.	Number of hills used.	Total number of shoots.	Total number of tubers.	Number of tubers per hill.	Number of tubers per shoot.	Total weight of tubers.	Grams.	Average weight of tubers.	Age of plant.	Benizidin.	Pyrogallol.	α -naphthol.	L. b. of m. g. ^a	Phloroglucin.	Alolin.	Pyrocatechol.	Tyrosin.	Hydrochinone.	Phloridzin.	Resorcin.	Guaiacol.	O-cresol.	M-cresol.	O-toluidin.	M-toluidin.	P-toluidin.	
46	21	Aug. 4	Aug. 13	2	4	13	6.5	3.2	114	8.8	62	0.121	0.137	0.070	0	0	0	0.281	0.682	0.281	0.082	0.257	0	0.218	0	0.371	0.515	0	0	0.043
47	49	Aug. 21	Aug. 22	5	32	101	20.2	3.2	565	5.6	40	0.265	0.218	0.117	0.016	0.047	0.023	0.367	0.140	0.367	0.140	0.367	0.281	0.047	0	0.371	0.515	0	0	0.043
48	32	Aug. 21	Aug. 22	5	32	101	20.2	3.2	565	5.6	40	0.265	0.218	0.117	0.016	0.047	0.023	0.367	0.140	0.367	0.140	0.367	0.281	0.047	0	0.371	0.515	0	0	0.043
49	50	Aug. 21	Aug. 22	5	32	101	20.2	3.2	565	5.6	40	0.265	0.218	0.117	0.016	0.047	0.023	0.367	0.140	0.367	0.140	0.367	0.281	0.047	0	0.371	0.515	0	0	0.043
50	51	Aug. 21	Aug. 22	5	32	101	20.2	3.2	565	5.6	40	0.265	0.218	0.117	0.016	0.047	0.023	0.367	0.140	0.367	0.140	0.367	0.281	0.047	0	0.371	0.515	0	0	0.043
51	34	Aug. 29	Aug. 30	1	1	5	5	5	470	94.0	88	0.203	0.031	0.023	0.020	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
52	35	Aug. 29	Aug. 30	1	1	5	5	5	470	94.0	88	0.203	0.031	0.023	0.020	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
53	54	Aug. 29	Aug. 30	1	1	5	5	5	470	94.0	88	0.203	0.031	0.023	0.020	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
54	55	Aug. 29	Aug. 30	1	1	5	5	5	470	94.0	88	0.203	0.031	0.023	0.020	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
55	56	Aug. 29	Aug. 30	1	1	5	5	5	470	94.0	88	0.203	0.031	0.023	0.020	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
56	57	Sept. 8	Sept. 9	1	1	4	4	4	240	60.0	98	0.179	0.191	0.172	0.008	0.016	0.047	0.226	0.086	0.386	0.242	0.008	0	0	0.406	0.831	0	0	0	

^a Leuco base of malachite green.

the other. The irregularities are due, no doubt, to differences between individual samples and to slight differences in the mode of preparation of the juice. Great care was used to maintain uniformity of technique throughout this work, so that it does not seem very likely that the latter factor plays a rôle in the variations of oxidase contents observed.

OXIDASE ACTIVITY OF THE JUICE OF SEED TUBERS

In connection with these results the oxidase activity of the seed tubers of the plants used in these experiments seemed of interest. The average weight of the tubers when examined was 90 grams. The oxidase determinations were started on July 18, when the tubers had just begun to sprout. The results are given in Table VIII.

TABLE VIII.—Oxidase activities of the juice of seed potato tubers from which all the healthy material used in this investigation was obtained

Series No.	Date.	Oxidase activity expressed in units as measured in the oxidation of the reagents.							
		Benzi- din.	Pyro- gallol.	a-naph- thol.	L. b. of m. g. ^a	Phloro- glucin.	Alcin.	Pyroca- techol.	Tyro- sin.
58.....	July 18								
59.....	July 18	0.179					0.031	0.261	0.109
60.....	July 19								
61.....	July 19								
62.....	July 19	0.226					0.055		
63.....	July 24					0.027			
64.....	July 24		0.254	0.125	0.086				

Series No.	Oxidase activity expressed in units as measured in the oxidation of the reagents.							
	Phlorid- zin.	Resorcin	Guaiacol.	Ortho- cresol.	Meta- cresol.	Para- cresol.	Ortho- toluidin.	Meta- toluidin.
58.....								
59.....			0.312					
60.....							0.008	0.000
61.....				0.047	0.530	0.913		
62.....			0.257					
63.....	0.359	0.005						
64.....								

^a Leuco base of malachite green.

OXIDASES OF CURLY-DWARF-DISEASED POTATO PLANTS

The plot of potatoes from which all the normal material was collected turned out to be remarkably free from curly dwarf. The pathological material was therefore collected on a larger field several miles remote from the other. About 10,000 different kinds of potatoes were grown in the field, and on account of the comparative scarcity of the pathological material samples were chosen from a number of different varieties. In all of the experiments the name of the variety is stated whenever it is known. The conditions of growth with reference to soil and atmospheric conditions were practically the same in the case of both the diseased and healthy potatoes.

TABLE IX.—Oxidase activities of the juice of shoots of curly-dwarf potato plants

Series No.	Date.	Number of variety.	Name of variety.	Number of plants.	Total weight of shoots.	Gms.	Mean total weight of shoots per hill.	Date of planting.	Age of plant.	Days.	Oxidase activity expressed in units as measured in the oxidation of the reagents.																				
											Benzidin.	Pyrogallol.	α -naphthol.	L. b. of m. g. a	Phloroglucin.	Alom.	Pyrocatechol.	Tyrosin.	Hydrochinone.	Phloridzin.	Resorcin.	Guaiacol.	O-cresol.	M-cresol.	P-cresol.	O-toluidin.	M-toluidin.	P-toluidin.			
65.....	July 17	Unknown.	Unknown.....	10	361	36	June 2	45	{	0.043					0.039	0.082	0.060	0.359			0.125										
66.....	July 21	do	do.....	10	435	44	do..	49	{	0.031					0.062			0.400			0.094	0.023	0.312	0.445							
67.....	July 22	do	do.....	7	260	37	do..	50	{																						
68.....	July 23	do	do.....	7	260	37	do..	51	{	0.074	0.023	0.047	0.031					0.220	0.047					0.047	0.057	0.117					
69.....	July 28	do	do.....	3	111	37	do..	56	{	0.062			0.016	0.020	0.030	0.031	0.047		0.374	0.187	0.043										
70.....	July 29	do	do.....	4	370	93	do..	57	{	0.062	0.055	0.016	0.023	0.023		0.078	0.023	0.452	0.371	0.031	0.078	0.039	0.140	0.382	0	0.012					
71.....	July 30	6652	Daisy X Round	5	150	30	do..	58	{	0.140	0.086	0.031	0.094	0.062	0.094		0.109	0.078	0.659	0.304	0.047	0.140									
72.....		4046	Prof. Maerker																												
73.....			X Silverskin.																												
74.....																															
75.....																															
76.....																															
77.....																															
78.....																															

a Leuco base of malachite green.

OXIDASE ACTIVITY OF THE JUICE OF THE SHOOTS

The procedure was very much like that used with healthy potato plants. The first experiments were carried out on the whole shoot of the plants. The results are summarized in Table IX.

There is no definite tendency observable in these results. The diluting influence of the stems is apparently more than compensated for by the

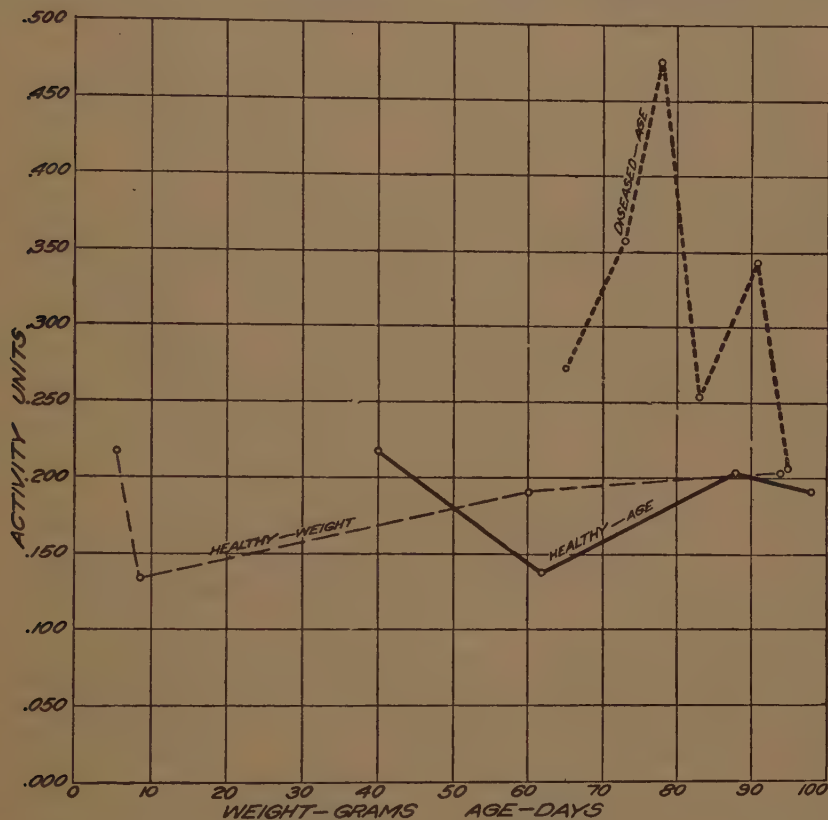


FIG. 13.—Curve showing oxidation of pyrogallol in the presence of the juice of potato tubers.

relatively higher oxidase content of the foliage of the older plants. All of the plants were started at the same time, so that from the viewpoint of age the results are comparable. As is shown by Table IX, no direct correlation can be found between the age or weight of the shoot and the oxidase content. These results are further discussed, together with other data on diseased foliage, on page 399.

OXIDASE ACTIVITY OF THE JUICE OF THE LEAVES

The results obtained in working with the foliage of curly-dwarf potato plants rather than with the whole shoots are given in Table X.

TABLE X.—*Oxidase activities of the juice of the leaves of curly-dwarf potato plants*

Series No.	Date.	No. of variety.	Number of hills used.	Total weight of shoots.	Gms.	Mean total weight of shoots per hill.	Number of shoots.	Mean weight of shoots.	Gms.	Date of planting.	Age of plant.	Total weight of stems.	Gms.	Stems $\times 100 =$ total weight of shoots.	Oxidase activity expressed in units as measured in the oxidation of the reagents.																																																									
															Benzidin.	Pyrogallol.	α -naphthol.	L. b. of m. & a.	Phloroglucin.	Alum.	Pyrocatechol.	Tyrosin.	Hydrochinone.	Phloridzin.	Resorcin.	Guaiacol.	O-cresol.	M-cresol.	P-cresol.	O-toluidin.	M-toluidin.	P-toluidin.																																								
79	Aug. 5	b ₁₄₉₈₀	4	380	95	4	95	4	95	June 2	64	178	47	0.062	0.039	0.047	0.055	0.070	0.070	0.374	0.234	0.069	0.078	0.031	0.211	0.371	0.031	0.055	0.078	0.031	0.055	0.078																																								
80																																	Aug. 6	c ₅₀₁₄	3	440	147	12	37	do....	65	254	58	0.109	0.047	0.047	0.086	0.086	0.086	0.086	0.250	0.039	0.055	0.047	0.265	0.398	0.020	0.047	0.062													
81																																	Aug. 7	d ₅₄₃₈	3	530	177	14	177	14	38	do....	66	284	54	0.031	0.035	0.031	0.008	0.035	0.016	0.051	0.307	0.226	0.027	0.078	0.047	0.226	0.320	0.031	0.035	0.053										
82	Aug. 6	c ₅₀₁₄	3	440	147	12	127	12	37	do....	65	254	58	0.109	0.047	0.047	0.086	0.086	0.086	0.250	0.039	0.055	0.047	0.265	0.398	0.020	0.047	0.062																																												
83																													Aug. 7	d ₅₄₃₈	3	530	177	14	177	14	38	do....	66	284	54	0.031	0.035	0.031	0.008	0.035	0.016	0.051	0.307	0.226	0.027	0.078	0.047	0.226	0.320	0.031	0.035	0.053														
84																													Aug. 8	e ₂₂₀₃	4	642	161	15	161	15	43	do....	73	315	49	0.101	0.043	0.055	0.051	0.059	0.117	0.047	0.585	0.261	0.055	0.137	0.047	0.312	0.460	0.078	0.140	0.055														
85	Aug. 19	4	500	125	20	25	do....	25	do....	78	198	40	0.039	0.016	0.039	0.016	0.031	0.101	0.043	0.406	0.290	0	0.105	0.047	0.261	0.390	0.043	0.031	0.055	0.055																																									
86																																Aug. 25	f ₆₀₆₁	2	890	445	5	178	do....	84	440	50	0.070	0	0.062	0.051	0.078	0.078	0.035	0.575	0.218	0.055	0.086	0.047	0.410	0.062	0.101	0.078	0.101	0.078												
87																																Aug. 27	g ₂₃₄₅₇	5	550	110	7	79	do....	86	210	38	0.016	0.047	0.047	0.113	0.105	0.113	0.078	0.068	0.347	0.125	0.047	0.410	0.062	0.101	0.078	0.101	0.078	0.101	0.078											
88	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																								
89																																	Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554										
90																																	Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554								
91	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																						
92																																			Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554						
93																																			Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554				
94	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																				
95																																					Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554		
96																																					Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554		
97	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																			
98																																						Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554	
99																																						Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554	
100	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																			
101																																						Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554	
102																																						Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554	
103	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																		
104																																							Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
105																																							Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
106	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																		
107																																							Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
108																																							Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
109	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																		
110																																							Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
111																																							Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
112	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554																																		
113																																							Sept. 2	i ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
114																																							Sept. 3	j ₁₈₇₄₃	4	440	110	7	63	do....	91	180	42	0.016	0.047	0.047	0.113	0.105	0.113	0.148	0.117	0.324	0.086	0.156	0.086	0.464	0.554
115	Sept. 1	h ₁₈₇₄₃	4	440	110	7	63	do....	63	do....	91	180	42	0.016	0.047																																																									

^a Early Eureka \times Keeper.
^b A. S. demissum \times Keeper.

^c Sophie \times Keeper.
^d Daisy \times Keeper.

^e Pres. Kruger \times Keeper.
^f Prof. Maerker \times Apollo.

^g Leuco base of malachite green.
^h Keeper \times Silverskin.

In order to ascertain whether the oxidase content of the leaves of these abnormal plants showed with age similar fluctuations to those found in healthy plants, curves representing the measurements of these oxidases were plotted with the curves representing the oxidase measure-

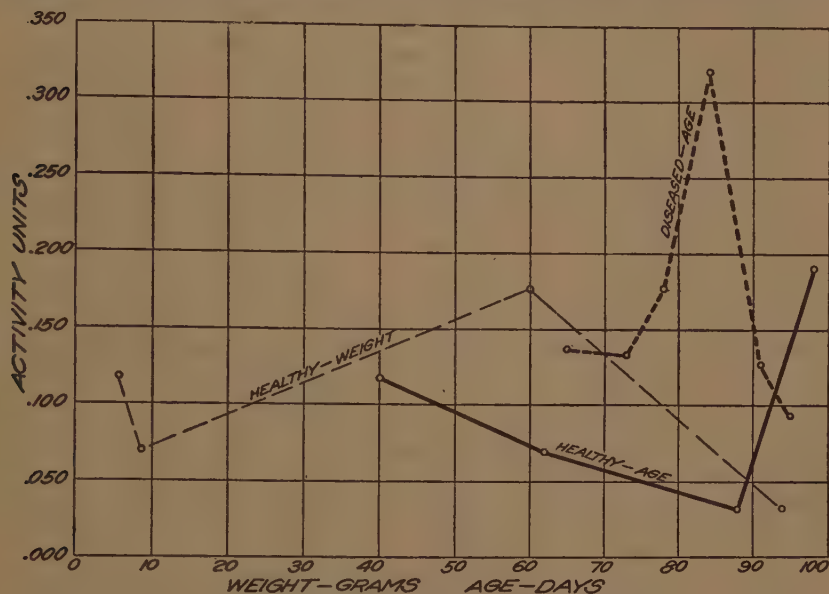


FIG. 14.—Curve showing oxidation of α -naphthol in the presence of the juice of potato tubers.

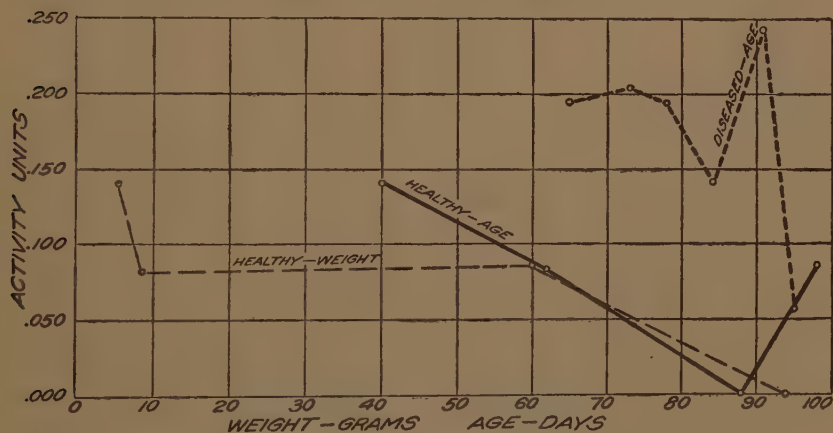


FIG. 15.—Curve showing oxidation of tyrosin in the presence of the juice of potato tubers.

ments of the healthy plants. Some of these are shown by the broken lines in figures 6 to 12. Plotting both curves on the same system of coordinates has the additional advantage of making possible a quick observation of the comparative magnitudes of oxidase activity in the healthy and diseased juices.

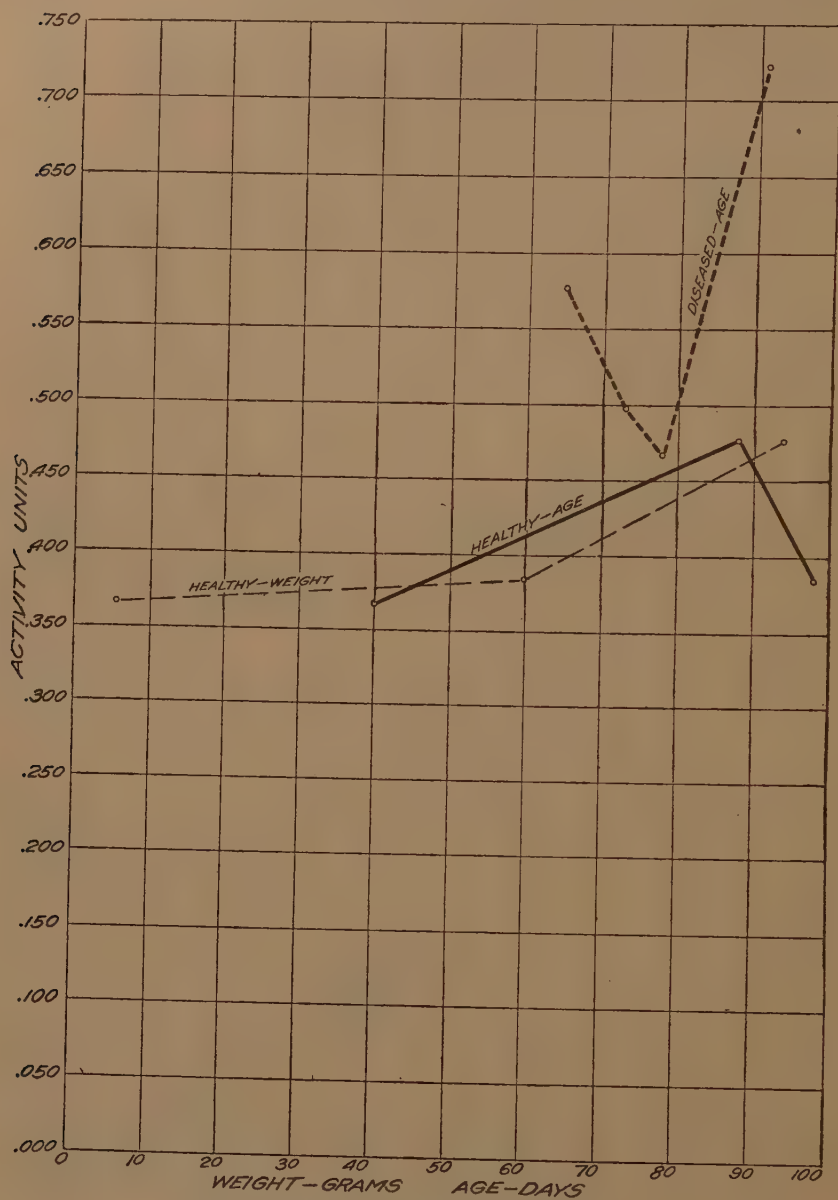


FIG. 16.—Curve showing oxidation of hydrochinone in the presence of the juice of potato tubers.

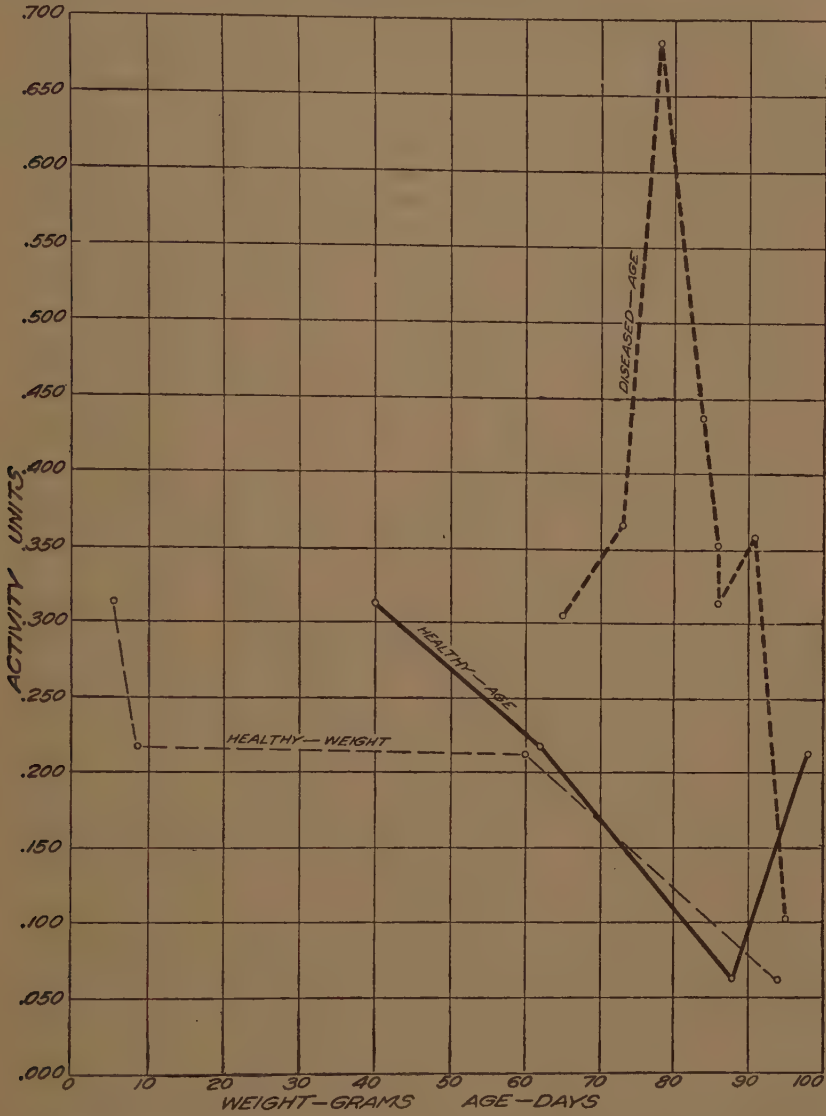


FIG. 17.—Curve showing oxidation of guaiacol in the presence of the juice of potato tubers.

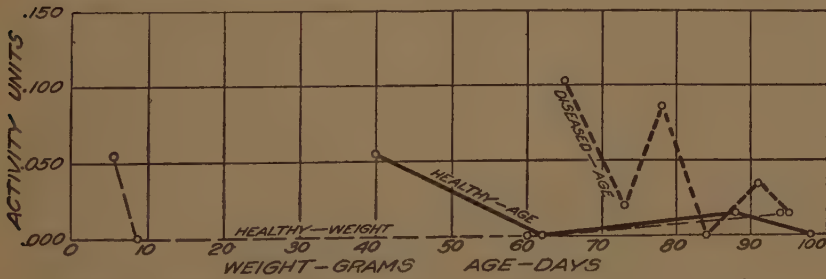


FIG. 18.—Curve showing oxidation of o-cresol in the presence of the juice of potato tubers.

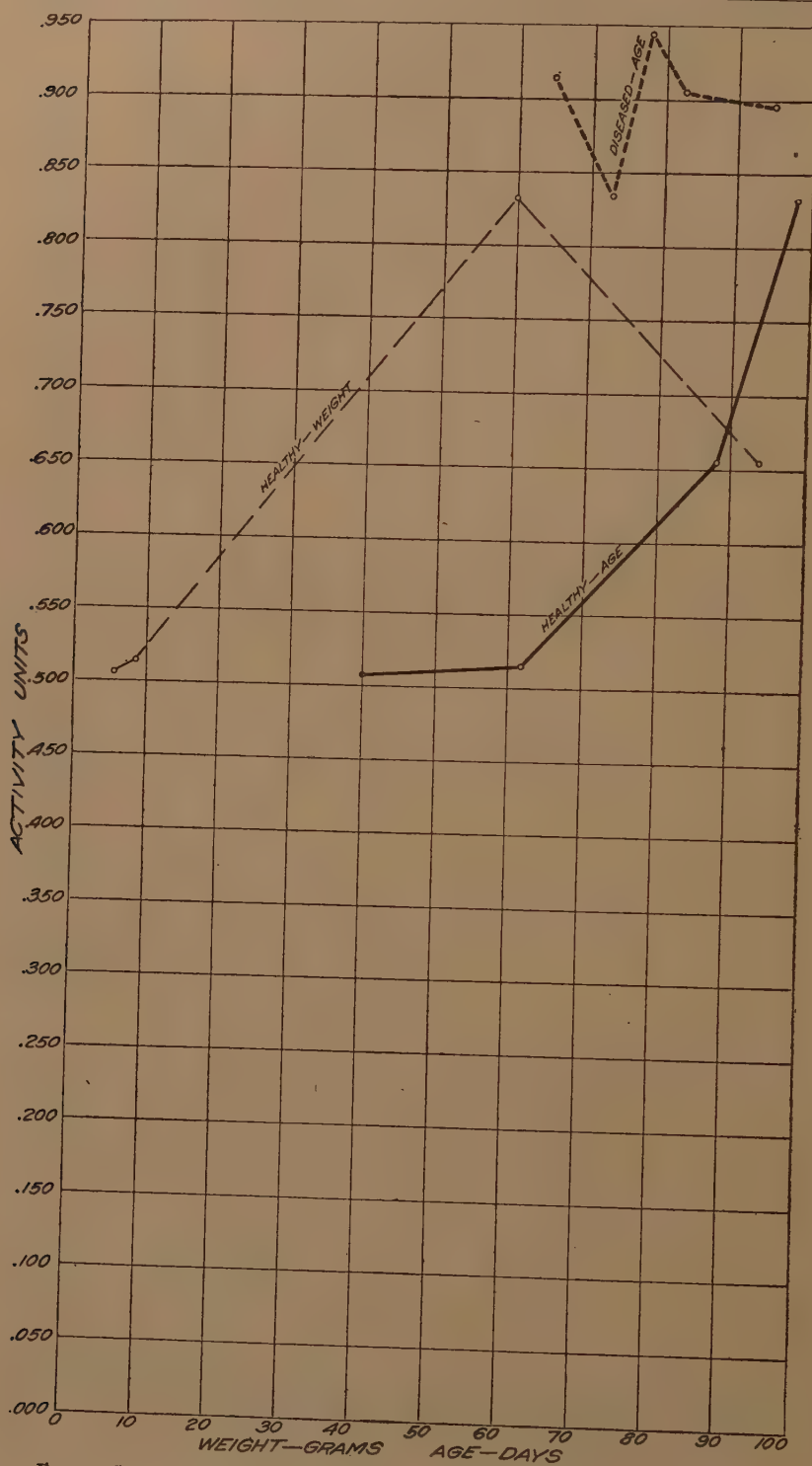


FIG. 19.—Curve showing oxidation of p-cresol in the presence of the juice of potato tubers.

These curves seem to show no definite tendency such as was seen in the case of the growing leaves of healthy plants. This is to be expected when it is considered that the physiological condition of the plants and presumably the oxidase contents of the juices are here influenced by two factors combined, age and disease. Past experience has shown that the oxidase activity of the plant juices is markedly affected by physiological disturbances such as the curly-dwarf disease of potatoes seems to be. The magnitude of the effect on the oxidase activities probably depends on such factors as the age of the plant when the disease first took hold,

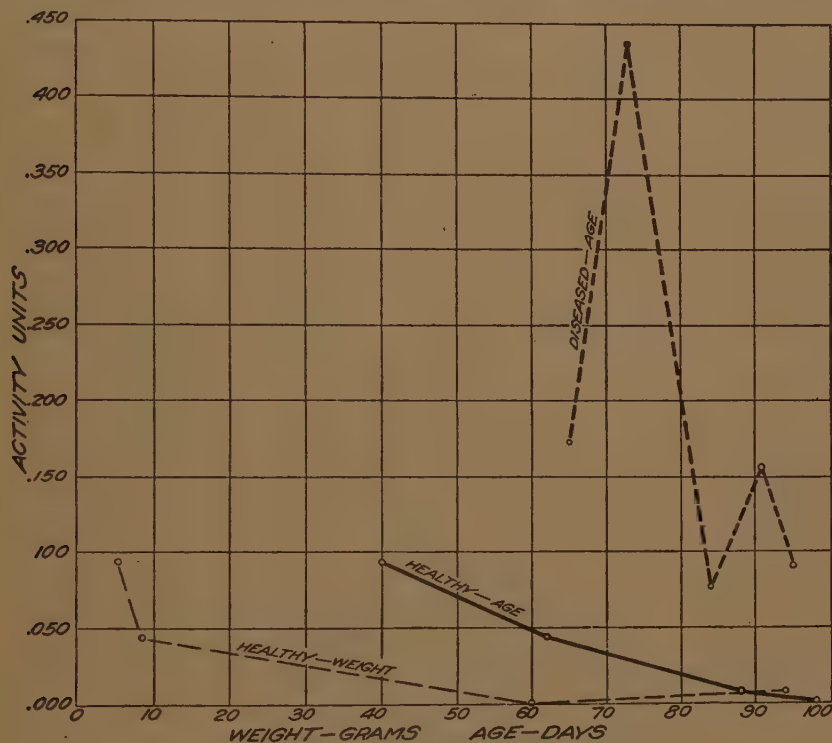


FIG. 20.—Curve showing oxidation of p-toluidin in the presence of the juice of potato tubers.

the length of time elapsed since, the individual resistance of the plant, etc. None of the factors are susceptible of measurement; the plants examined are influenced by them to different degrees and are therefore not comparable.

However, they all showed the typical curly-dwarf symptoms, and if the oxidase activities of the leaf juice are influenced by the apparent physiological disturbances the influence should be noticeable by a deviation of the oxidase activities from the normal and in a definite direction. That such a deviation from the normal actually exists is indicated by the curves in figures 6 to 12. With most of the reagents used the broken

lines, representing the oxidase activities of the curly-dwarf foliage, run at a higher mean level than the continuous lines, which represent the oxidase activities of the healthy foliage. The differences will be brought out in a mathematical form in a latter part of this paper.

To get a clear idea of the striking differences in the rate and extent of growth existing between the healthy and the diseased plants, the results showing these differences are represented graphically in figure 21. The ages of the plants are represented on the abscissæ, the mean weight

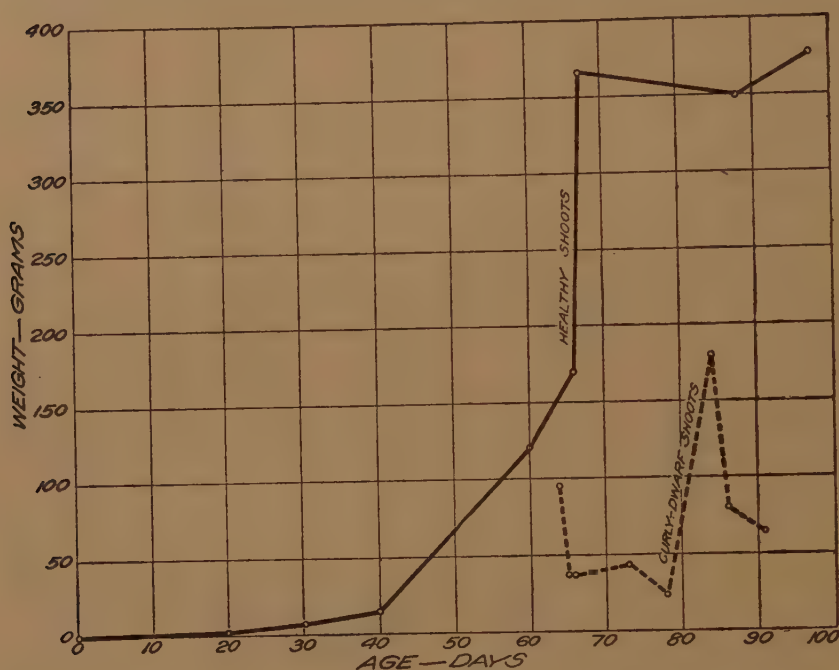


FIG. 21.—Curve showing the differences in rate and extent of growth between healthy and diseased plants.

of the shoots on the ordinates. The continuous line represents the growth of the healthy plants, the broken line that of the diseased ones.

The difference in weight of the two types of potato plants of the same age is strikingly apparent. The diseased plants made an average growth of only about one-eighth of the growth of the normal plants.

The fluctuations in the curve representing the rate of growth of curly-dwarf shoots are to be expected when the complex nature of the disease is considered. The diseased plants used for experimentation, although showing the typical symptoms, differed greatly in size, as is shown in figure 21, in color, and also in the shape which they assumed on account of the inhibition of growth.

OXIDASE ACTIVITY OF THE JUICE OF THE TUBERS

The collection of the tubers of the diseased plants and the determinations of the oxidase content of the juices obtained from them were carried out in the same way as was done in the case of the healthy material. The results are summarized in Table XI and are included in figures 13 to 20. As before, the heavy broken lines represent the results obtained with curly-dwarf material.

These results, like those obtained with the tubers of healthy potato plants, show no definite tendency. If with age there is a definite variation in oxidative capacity exhibited toward all of the reagents, it is entirely masked by the irregular fluctuations. These irregular fluctuations were also observed in the case of diseased foliage and are illustrated in figures 6 to 12.

DISCUSSION OF RESULTS

Comparison of the curves of the healthy plants with those of the diseased ones shows at a glance a greater oxidase activity in the case of the curly-dwarf material. This is true for both the tubers and the foliage. It seemed desirable to express these differences in some numerical form, and this was done by taking the averages of all the results obtained from material of the same type with the same reagents. These averages were then easily compared.

It was shown that healthy foliage yields juices of diminishing oxidase activity from the time of sprouting up to about the fortieth day of growth (as counted from the time of planting). For this reason in this summary of averages must be included only those of the results obtained with healthy leaves which were obtained during the growth periods of the diseased material examined. The age of the diseased foliage collected ranged from 64 to 91 days; the age of the plants where the whole shoots were examined was from 45 to 58 days. The averages were calculated as follows: All of the data (oxidase activities) obtained within the age periods mentioned were added together with the figures obtained for the beginning and the end of the period by interpolation from the curve. The sum, of course, was divided by the number of data added. These averages are shown in Table XII.

51131°—14—6

TABLE XI.—Oxidase activities of the juice of potato tubers

Series No.	Series No. of leaves of same.	Date of collection.	Date of experiment.	Number of hills used.	Total number of shoots in same.	Total number of tubers.	Number of tubers per hill.	Number of tubers per shoot.	Total weight of tubers.	Gms.,	Mean weight of tubers.	Age of plant.	Oxidase activity of juice of tubers expressed in units using the reagents.																	
													Benzidin.	Pyrogallol.	<i>o</i> -naphthol.	L. b. of m. R. ^a	Phloroglucin.	Alcin.	Pyrocatechol.	Tyrosin.	Hydrochinone.	Phloridzin.	Resorcin.	Guaiacol.	O-cresol.	M-cresol.	P-cresol.	O-toluidin.	M-toluidin.	P-toluidin.
102	82	Aug. 6	Aug. 12	3	12	11	3-7	0-9	245	Gms.,	22-3	65	0-413	0-273	0-137	0-059	0-035	0-047	0-435	0-195	0-577	0-211	0	0-304	0-101	0-647	0-913	0-004	0-031	0-172
103	104																													
104	105	Aug. 14	Aug. 15	4	13	27	6-8	1-8	175	Gms.,	6-5	73	0-359	0-133	0-031	0-023	0-023	0-023	0-82	0-203	0-499	0-234	0-008	0-307	0-020	0-515	0-835	0	0-031	0-031
105	106																													
106	107																													
107	108																													
108	109	Aug. 19	Aug. 20	4	20	94	23-5	4-7	365	Gms.,	3-9	78	0-408	0-179	0-023	0-070	0-070	0-070	0-472	0-195	0-468	0-257	0-686	0-593	0-944	0-008	0-008	0-437	0-031	0-031
109	110																													
110	111																													
111	112																													
112	113	Aug. 25	Aug. 26	2	5	15	7-5	3-0	530	Gms.,	35-3	84	0-250	0-254	0-020	0-031	0-031	0-031	0-281	0-140	0-289	0-289	0-437	0-886	0-593	0-944	0-008	0-008	0-437	
113	114																													
114	115																													
115	116	Aug. 27	Aug. 28	5	7	14	2-8	2-0	505	Gms.,	36-0	86	0-086	0-343	0-129	0-023	0-023	0-023	0-316	0-205	0-234	0-234	0-312	0	0-218	0-905	0	0	0-078	
116	117																													
117	118																													
118	119	Sept. 1	Sept. 2	4	7	17	4-3	2-4	265	Gms.,	15-6	91	0-086	0-343	0-129	0-023	0-023	0-023	0-078	0-242	0-722	0-254	0-359	0-035	0-460	0	0	0	0-156	
119	120																													
120	121	Sept. 5	Sept. 6	3	3	33	11	590	Gms.,	17-9	95	0-176	0-207	0-094	0-016	0-031	0-043	0-059	0-059	0-195	0-016	0-101	0-016	0-897	0	0	0	0-090	
121	(b)	Sept. 5	Sept. 6	3	3	33	11	590	Gms.,	17-9	95	0-176	0-207	0-094	0-016	0-031	0-043	0-059	0-059	0-195	0-016	0-101	0-016	0-897	0	0	0	0-090	

^a Leuco base of malachite green.

^b Foliage dead.

^a Leuco base of malachite green.^b Foliage dead.

TABLE XII.—*Relative oxidase activity of healthy and curly-dwarf diseased potatoes*

Reagent used.	Oxidase activity ^a of juices of—						Difference in activity between juices of—		
	Shoots of healthy plants collected at the age of 43 to 58 days.	Shoots of curly-dwarf plants collected at the age of 45 to 58 days.	Foliage of healthy plants collected at the age of 64 to 91 days.	Foliage of curly-dwarf plants collected at the age of 64 to 91 days.	Tubers of healthy plants.	Tubers of curly-dwarf plants.	Curly-dwarf and healthy shoots.	Curly-dwarf and healthy foliage.	Curly-dwarf and healthy tubers.
Benzidin.....	0.011	0.085	0.062	0.061	0.188	0.231	Per cent. + 673	Per cent. + 54	Per cent. + 23
Pyrogallol.....	0.002	0.053	0.026	0.040	0.187	0.318	+ 2,550	+ 70	+ 168
α-naphthol.....	0.011	0.027	0.008	0.025	0.105	0.098	+ 145	+ 213	+ 47
Leuco base of malachite green.....	0.014	0.043	0.040	0.057	0.015	0.022	+ 207	+ 42	+ 26
Phloroglucin.....	0.017	0.034	0.045	0.045	0.023	0.029	+ 100	—	+ 39
Aluin.....	0.013	0.048	0.005	0.056	0.023	0.032	+ 309	+ 14	+ 54
Pyrocatechol.....	0.027	0.076	0.056	0.086	0.261	0.268	+ 181	+ 54	+ 123
Tyrosin.....	0.016	0.057	0.068	0.064	0.077	0.172	+ 250	+ 8	+ 3
Hydrochinone.....	0.312	0.450	0.382	0.450	0.409	0.507	+ 44	+ 18	+ 39
Phloridzin.....	0.112	0.272	0.106	0.269	0.248	0.243	+ 143	+ 154	+ 2
Resorcin.....	0.023	0.042	0.058	0.046	0.016	0.004	+ 83	+ 22	+ 75
Guaiacol.....	0.047	0.103	0.079	0.102	0.200	0.304	+ 119	+ 29	+ 82
O-cresol.....	0.041	0.031	0.045	0.050	0.018	0.043	+ 11	+ 11	+ 139
m-cresol.....	0.169	0.226	0.108	0.290	0.388	0.487	+ 34	+ 103	+ 45
p-cresol.....	0.266	0.414	0.203	0.414	0.627	0.919	+ 50	+ 168	+ 47
O-toluidin.....	0.036	0.024	0.057	0.044	0	0.002	+ 33	+ 23	...
m-toluidin.....	0.040	0.029	0.060	0.068	0.008	0.012	+ 28	+ 13	+ 50
p-toluidin.....	0.028	0.065	0.052	0.059	0.036	0.187	+ 132	+ 13	+ 419

^a Activity expressed in units as measured in the oxidation of the reagents.

As Table XII shows, the differences existing between the oxidase activity of the healthy and of the diseased material are generally marked and the greater activity is in the curly-dwarf potato plants. The comparison of the data for healthy and curly-dwarf shoots shows that among the 18 reagents only 3 are oxidized more readily in the presence of the juice of the healthy plants. Comparison of the leaves of the two types of plants shows 7 of the 18 reagents to be more readily oxidized by the healthy juice; in the case of the two types of tubers only two of the reagents showed greater oxidation by the healthy material. Among 54 sets compared, 12 showed a greater activity in the case of the healthy material, while the remainder, 42, showed a much greater activity in the case of the diseased plants.

It seems safe to conclude that in general the oxidizing power in the juices of curly-dwarf potato plants is greater than in those of healthy plants. The writer does not know as yet exactly what bearing, if any, the oxidases measured by him have on the oxidation processes going on in the cells. *A priori*, one would conclude that the intensity of oxidation processes in the cells would among other factors depend on the concentration of the various oxidases present. If this were the case, one would expect cell respiration to be more intense in the cells of the curly-dwarf tubers. The diseased plants would be in a condition corresponding to "fever" in animals.

These results agree in their general nature with those obtained in the case of the curly-top of sugar beets (Bunzel, 1913a, 1913b) and the leaf-roll of potatoes (Doby, 1911-12). In all three cases an increase in oxidases and a general retardation of growth are found. It would be extremely interesting, especially to plant physiologists, to find out what the rate of respiration is in such dwarfed, presumably "feverish" plants. Experiments intended to throw light on this point are already being planned in the laboratory of the Office of Plant Physiological and Fermentation Investigations.

There are a number of facts brought out in this investigation which open doors to new aspects of the physiology of development. There seems to be a cycle in the activity of the expressed juice of the foliage of normally developing potato plants. The juice of the foliage of very young plants is more active than that of plants of the same variety 40 or 50 days older; after that stage of development the activity rises again with increasing age. Quite in harmony with these findings is the fact that sprouts of artificially sprouted tubers of the same variety are much more active than the youngest foliage examined.

There seems to be a parallelism, therefore, between the intensity of physiological activity and the quantity of oxidases present. This belief is strongly corroborated by the fact that the physiologically more active portions of the plant, such as the leaves, furnish juices with greater activity than the obviously less active portions of the same plant, such as

the stems. This has been found by the writer not only in the case of the potato plants, but also in sugar beets (Bunzel, 1913a, 1913b).

In this connection the results obtained by Nicolas (1907) are very interesting. He studied the respiration of individual parts of plants and found that those organs which carry out the assimilating functions of the plant show the greatest respiratory activity. The limbs or the organs which replace them in function, such as the phyllodia or cladodia, have 1.4 to 4.5 times as great a respiratory activity as the petiole, stem, or tendrils. These results when combined with those obtained by the writer in the present investigation would indicate that there is at least a general parallelism between the oxidase activity of the juice obtained from a plant organ and the intensity of its physiological activity, as measured by its intensity of respiration. Plans are made to study the question more closely in the laboratory of the Office of Plant Physiological and Fermentation Investigations.

SUMMARY

(1) The oxidase activity of the foliage of normally developing potato plants is greatest in the early stages of development; it falls off with growth of the plants and rises again when the plant's growth about reaches a standstill.

(2) Curly-dwarf potato plants show a greater oxidase activity than healthy ones of the same age, both in the juice of their tubers and in the juice of their foliage.

(3) The oxidative activity of the different parts of the potato plant has been established for 18 different reagents.

LITERATURE CITED

BACH, A., and CHODAT, R.

1904. Untersuchungen über die Rolle der Peroxyde in der Chemie der lebenden Zelle. *In* Ber. Deut. Chem. Gesell., Jahrg. 37, no. 10, p. 2434-2440, 2 fig.

BERTRAND, GABRIEL.

1897. Sur l'intervention du manganèse dans les oxydations provoquées par la laccase. *In* Compt. Rend. Acad. Sci. [Paris], t. 124, no. 19, p. 1032-1035.

BUNZEL, H. H.

1912. The measurement of the oxidase content of plant juices. U. S. Dept. Agr. Bur. Plant Indus. Bul. 238, 40 p., 9 fig., 2 pl.

1913a. A biochemical study of the curly-top of sugar beets. U. S. Dept. Agr. Bur. Plant Indus. Bul. 277, 28 p.

1913b. Die Rolle der Oxydasen in der Blattrollkrankheit der Zuckerrübe. *In* Biochem. Ztschr., Bd. 50, Heft 3/4, p. 185-208.

DIXON, H. H., and ATKINS, W. R. G.

1913. Osmotic pressures in plants. I. Methods of extracting sap from plant organs. *In* Sci. Proc. Roy. Dublin Soc., n. s. v. 13, no. 28, p. 422-432.

DOBY, GÉZA.

1911-12. Biochemische Untersuchungen über die Blattrollkrankheit der Kartoffel. *In* Ztschr. Pflanzenkrank., Bd. 21, Heft 1/2, p. 10-17, Heft 6, p. 321-336, 1911; Bd. 22, Heft 4, p. 204-211, Heft 7, p. 401-403, 1912.

DONY-HÉNAULT, OCT.

1908. Contribution à l'étude méthodique des oxydases. *In Acad. Roy. Belg. Bul. Cl. Sci.*, 1908, no. 2, p. 105-107.

—— and VAN DUUREN, J.

1907. Contribution à l'étude méthodique des oxydases dans les tissus animaux. *In Acad. Roy. Belg. Bul. Cl. Sci.*, 1907, no. 5, p. 537-638.

GRÜSS, J.

1907. Abhandlungen über Enzymwirkungen. *In Ztschr. Pflanzenkrank.*, Bd. 17, Heft 2, p. 65-79, 1 fig., pl. 4.

KÖNIG, J., and KRÜSS, H.

1904. Erläuterungen zur Feststellung des Trübungsgrades und der Farbentiefe von Flüssigkeiten mittels des Nephelometers. *In Ztschr. Untersuch. Nahr. u. Genussmtl.*, Bd. 7, Heft 10, p. 587-590.

NICOLAS, G.

1907. Sur la respiration des organes végétatifs aériens des plantes vasculaires. *In Compt. Rend. Acad. Sci. [Paris]*, t. 144, no. 20, p. 1128-1130.

ORTON, W. A.

1914. Potato wilt, leaf-roll, and related diseases. *U. S. Dept. Agr. Bul.* 64, 48 p., 16 pl. Bibliography, p. 44-48.

PALLADIN, W.

1906. Bildung der verschiedenen Atmungsenzyme in Abhängigkeit von dem Entwicklungsstadium der Pflanzen. *In Ber. Deut. Bot. Gesell.*, Bd. 24, Heft 2, p. 97-107, pl. 8.

SORÄUER, PAUL.

1908. Die angebliche Kartoffelepidemie, genannt die "Blattrollkrankheit." *In Internat. Phytopath. Dienst (Beigabe, Ztschr. Pflanzenkrank.)*, Jahrg. 1, Stück 2, p. 33-59.

TRILLAT, A.

1903. Influences activantes ou paralysantes agissant sur le manganèse envisagé comme ferment métallique. *In Compt. Rend. Acad. Sci. [Paris]*, t. 137, no. 22, p. 922-924.

1904. Sur le rôle d'oxydases que peuvent jouer les sels manganeux en présence d'un colloïde. *In Compt. Rend. Acad. Sci. [Paris]*, t. 138, no. 5, p. 274-277.



